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COLLEGE OF AGRICULTURAL AND
ENVIRONMENTAL SCIENCES
AGRICULTURAL EXPERIMENT STATION
COOPERATIVE EXTENSION

DEPARTMENT OF WILDLIFE, FISH, AND CONSERVATION BIOLOGY
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7-26-97

CALFED Proposal Reviewers
Sacramento, CA

Dear Reviewers:

I enthusiastically support the delta smelt rearing proposal of Dr. Doroshov and Dr. Lindberg. They plan to test a variety of factors considered important in the rearing success of this fish. In small scale experiments, they will examine effects of water chemistry and the presence of algae on the rotifer feeding response in the youngest (feeding) life history stages of delta smelt. They also plan to test temperature effects on larval and juvenile growth. These experiments should lead to improved delta smelt production capabilities by the third year of the study.

Many of us recognize the need for delta smelt. The reared fish would be very useful for toxicity testing, for experiments with fish screening devices (such as those that my colleagues and I have proposed to CALFED), and potentially for testing habitat suitability or for restocking parts of the Sacramento - San Joaquin Delta estuary.

The proposed project would be a cooperative effort between California and federal agencies to surmount the final hurdles regarding the mass production of delta smelt. I fully support this effort.

Sincerely,

Joseph J. Cech, Jr.
Professor of Fisheries Biology

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DWR

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RE: Calfed grant proposal

Dear Dr. Doroshov,

We were delighted to learn that you are submitting a proposal to Calfed on laboratory breeding and culture of delta smelt for developmental and environmental studies. As stated in our proposals to Calfed (Bennett, Teh , and Anderson) and IEP (Teh and Hinton), we are requiring laboratory-raised delta smelt for ambient water toxicity testing and for determination of responses to exposure. We will compare the laboratory-exposed fish to field-collected fish and correlate cause and effect of contaminants in the Delta ecosystem. Furthermore, due to the fact that there is no delta smelt control site in California, your proposal is very critical to all investigators working on contaminant effects in delta smelt. The funding of your study will provide a unique interdisciplinary, lab-field approach not otherwise available.
We strongly support your proposal and will be glad to collaborate with you on your developmental studies.

Sincerely,
David E. Hinton Swee Teh

Swee J. Teh and David E. Hinton (Director)
Aquatic Toxicology Program
University of California-Davis

F1-166

CULTURE OF DELTA SMELT, *Hypomesus transpacificus*, AT DELTA SITE, IN SUPPORT OF ENVIRONMENTAL STUDIES AND RESTORATION

Prof. Serge Doroshov, Dr. Joan Lindberg,
Joel Van Eenenaam, and Brent Baskerville-Bridges
University of California-Davis

I. Executive Summary

I. b. Project Description and Primary Biological/Ecological Objectives

The goal of the three-year project is to establish a functional culture system at a Delta site for the threatened delta smelt. Such a system, based on the annual procurement of 200-300 wild brood fish, will support ongoing environmental projects by providing all life stages of smelt for laboratory research. To achieve this goal, we will design and operate two small culture facilities, and optimize the methodologies of culture at these facilities, the State Water Project (SWP) Fish Facility and Fish Rearing Facility at the Federal Station in Byron. The main objectives of the project are: (1) to develop a reliable and technically feasible culture system encompassing all life stages of delta smelt; (2) to establish and characterize culture methods, environmental and technical parameters of culture system, and its production capabilities; (3) to initiate the supply of live material for testing in research laboratories, as well as preserved material and data that can be used as test-standards in ongoing projects of delta smelt habitat improvement.

I. c. Approach/Tasks/Schedule

The approach to culture system is based on our previous smelt projects at the SWP in Byron (Lindberg 1992, 1996) and at UC Davis (Mager et al. 1996), and collaboration with two laboratories at UC Davis (Fish Diseases, Dr. R. Hedrick and Fish Physiology, Dr. J. Cech). Delta smelt is an annual species maturing at the end of first year of life and spawning in spring. With relatively small egg yolk reserves, their newly emerged pelagic larvae start exogenous feeding on the fifth day after hatching. However, the duration of the larval stage to metamorphosis is extended for almost 3 months necessitating utilization of cultured and wild zooplankton for feeding larvae in culture. Our culture program will follow, with modifications, five basic steps: the procurement of wild juveniles, rearing them to spawning in flow-through tanks, hatching embryos in jars, rearing larvae to post-larval stage on cultured rotifers, and rearing post-larvae to full metamorphosis on natural and cultured zooplankton. Moving the entire life cycle culture system to site in the delta will provide advantages of improved spawning performance and availability of the natural zooplankton for rearing larvae to metamorphosis. The schedule and tasks of the project follow:

Year 1. a) installation of rearing system for the broodstock and culture unit for phytoplankton and rotifers at the Federal Station; b) rearing and spawning brood fish at both sites; c) optimization of larval culture (use of phytoplankton); d) rearing wild post-larval stages of delta smelt trapped at the Fish Collection Facilities, State and Federal.
Year 2. a) optimization of temperature and feeding regimes in larval culture; b) preliminary evaluation of larval survival, development, and growth.
Year 3. a) adjustments in production system based on the results of two years; b) evaluation of performance and production capabilities of culture system; c) summary evaluation of system design, culture protocols, and methodologies.
We will supply research laboratories and agencies with delta smelt of different life stages. Our target parameters for the culture system utilizing 250 brood fish (with minimum 100 females) are

35,000 fertilized eggs, 28,000 late-stage embryos or newly emerged larvae, and 10,000 juveniles at metamorphosis. The production system is expected to be compact but labor-intensive. The culture system can be used as a prototype for other species with pelagic larvae, such as the longfin smelt, american shad, and striped bass.

I. d. Justification for Project and Funding by CALFED

The delta smelt population and its unique upper estuary habitat are affected by a variety of environmental changes in ecosystem. There are ongoing projects supported by CALFED on habitat restoration and improvement in the Delta. Current supply of live delta smelt for the laboratory studies is scarce, unpredictable, and limited to adult/subadult life stages whereas the major impact may occur on early life stages. The proposed project will support ongoing studies on fish screen design, environmental physiology, eco-toxicology, and habitat improvement by providing smelt embryos, larvae, juveniles, and adults for laboratory testing. Additionally, preserved specimens and development growth-charts can be used as the standard for the evaluation of fish performance in the wild, and restored habitats.

I. e. Budget Costs

Project annual cost (includes 10% indirect cost) is $194,870 for the year 1, $195,537 for the year 2, and $202,369 for the year 3. The major part of the budget supports three key personnel who will be working full time at delta smelt culture facility. Their previous experience and technical skill are important for culture of delta smelt.

I. f. Applicant Qualifications

Dr. Serge Doroshov has research experience and expertise in developmental biology and hatchery technology of cultured fish, including sturgeon, striped bass, catfish, trout, and marine species. He and his graduate student, Dr. Randy Mager, have developed prototype culture-system for delta smelt at UC Davis and characterized sexual maturation, gametogenesis, and early development in this species. Dr. Joan Lindberg conducted her graduate studies on salmon metamorphosis and feeding behavior in sturgeon larvae. She led an independent project on delta smelt culture at SWP facilities in Byron before joining the university team. Joel Van Eenennaam has vast experience in breeding and culture of various fish species; he gained national reputation as one of the best aquacultural researchers. Brent Bridges conducted his M.S. project on larval nutrition and has excellent expertise in algal-rotifer production for larval culture, including 2 years experience with delta smelt. Jennie Kulczyk has completed a BS Degree in Biology and has developed experience in spawning, and rearing of larval and juvenile smelt at the SWP site.

I. g. Monitoring and Data Evaluation

Some of material of this project can be used as the standards for bio-monitoring program. For example, developmental charts for delta smelt can be used in the analysis of captured embryos, larvae, and post-larvae of smelt in different locations, to examine larval dispersal, growth and development in wild population.

I. h. Local Support/Coordination with other Programs/ Compatibility with CALFED objectives

The location of this project on site in the Delta is expected to enhance collaboration between State and Federal Agencies and university researchers. We will collaborate with the several laboratories and researchers at UC Davis.

CULTURE OF DELTA SMELT, *Hypomesus transpacificus*, AT DELTA SITE, IN SUPPORT OF ENVIRONMENTAL STUDIES AND RESTORATION

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A PROPOSAL TO THE CALFED BAY–DELTA PROGRAM

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Participants
California Department of Water Resources
Federal Bureau of Reclamation

RFP Project GroupType 3 (Services)

July, 1997

III. a. Project Description and Approach

The proposed delta smelt culture project comprises several objectives: (1) to develop and establish a reliable fish culture system for all life stages of delta smelt; (2) to characterize system parameters, methodologies, and production capabilities of the system, and (3) to initiate a supply of live material, at all life stages, for research projects concerned with a range of bay-delta fish issues. As a secondary objective we wish to test the effectiveness of holding and rearing wild caught 20 to 30-mm post-larvae. Capture of larvae from the wild has some risk (uncertainty, disease, and mortalities), but the potential salvage of post-larval smelt warrants testing. Since hatchery fish introduced into the wild can have a negative impact on natural populations, the proposed project will not restock delta smelt.

Our proposed project will build upon successes in delta smelt culture achieved over the last several years at the UC-Davis and the State Water Project (SWP) Fish Facility in Byron. Our team has successfully advanced methods for the capture, spawning, incubation, and rearing of smelt (Lindberg 1992, 1996; Mager 1996, Mager et al.1996). We propose to refine and apply these methods to the culture of delta smelt in the existing small laboratory at the State Water Project Fish Facility, and in the new fish-holding building at the nearby Federal pumping facility. We will obtain post-larvae for our second objective from fish salvage operations at the pumping facilities or from light traps set at the same sites.

The proposed supply of captive smelt will serve a variety of research interests. Smelt reared from laboratory embryos in clean well-water will be provided to toxicologists, who need fish reared in a clean environment. Fish culture is the only method of producing these fish. Other ongoing studies, such as fish screen development efforts, could be supplied with either hatchery-reared or captured juveniles.

III. b. Location

Two adjacent sites in the south delta will be used to develop and refine the delta smelt culture methods: an existing small laboratory at the SWP Fish Facility, and a new fish-holding facility at the nearby Federal pumping facility. The UC-Davis fish laboratories at the Institute of Ecology and Animal Science will be used for fish and tissue sample processing, water quality and data analysis.

III. c. Expected Benefits

The limited number of smelt in the wild and its threatened status, precludes obtaining large numbers of these animals from the wild for research. Furthermore the techniques for capture and holding of these fish have only been developed for the sub-adult to adult stages. Therefore the primary benefit of the proposed research is to create a supply of high quality material at all life stages (gametes, embryos, larvae, juveniles and adults) in order to advance research on smelt. The ready supply of animals will, in turn, accelerate research efforts aimed at determining the environmental factors impacting the smelt population.

Several existing and proposed projects have been identified that need smelt of various life stages to validate their work, or to proceed with their work. The research is aimed at creating a better understanding of smelt dispersal in juveniles (L. Grimaldo, DWR, and

D. Sweetnam, DFG, personal communication) and assessing the health of juvenile smelt at various delta locations - habitat and stressor evaluation (W. Bennett, and Swee Teh, UC-Davis, personal communication). Additionally, state and federal researchers are testing new fish screen designs, and these experiments require large numbers of smelt.

III. d. Background and Technical Justification

The decline in delta smelt abundance in the delta since the early 80's prompted listing the fish as *threatened* in the early 90's, and launched studies to determine the cause of the decline. Suggested causes include loss of shallow water habitat, entrainment at Federal and State pumping plants, competition with introduced species, contaminant concentration in the delta, and changes in prey organisms and abundance (Moyle et al., 1992; USFWS, 1995). Current research has begun to address the importance of these factors. Substantial resources are going into the design and reclamation of farm land and seasonally flooded lands to create spawning and nursery habitat for the delta smelt and other resident species. Other studies are assessing the health of smelt juveniles from various locations in the delta to understand the effect of variability in rearing habitat (and possible toxicity) as it impacts smelt health and growth (W. Bennett, UC-Davis, personal communication). Testing of smelt in the laboratory has provided information on temperature and salinity tolerances (Swanson and Cech, 1995) and swimming performance (Swanson and Cech, 1994). Efforts to refine fish-screen design are on-going at the State and Federal pumping facilities; due to the smelt's high sensitivity to many stressors, it is the primary test species for such studies. The limited number of smelt in the wild and its threatened status, precludes obtaining large numbers from the wild for this type of research.

Delta smelt culture should be based on scientific knowledge of the reproduction and development of the species—knowledge that has been largely unavailable until recent years. Two pilot scale studies were initiated in 1991 to begin characterizing the development of smelt (Mager, 1996), and to develop methodologies for the capture, handling, and rearing of these fish (Lindberg, 1992 and Mager et al., 1996). A summary of the delta smelt life cycle, as it was observed in the laboratory culture system, is shown in Figure 1. Adults (the great majority in the wild are apparently one-year-old annual fish) undergo gonadal recrudescence (meiosis in males and vitellogenesis in females) during a relatively short period of time (3-4 months) in the winter and early spring. An average female sheds about 1,000 adhesive eggs ranging from 0.8-1.0 mm in diameter. Hatching takes place 9-12 days after fertilization depending on temperature. Newly emerged larvae are small, transparent, and pelagic, inhabiting the "mixing zone" of the upper estuary; their yolk sacs are resorbed rapidly and they initiate exogenous feeding on the 5th day after hatching using primarily microplankton (cultured rotifers, 150-200 μm in size). Feeding, finfold larvae continue their passive pelagic life for a significant period of time, likely for two months, before completing differentiation of fins, inflation of swim bladders, and acquisition of hydrostatic regulation and improved swimming performance. Full metamorphosis (body remodeling and loss of tissue transparency) takes place at approximately 3 month of age and at a body size of 22-30 mm. The long duration of the larval stage in delta smelt is associated with larval dispersal strategy in this estuarine species. It is a significant challenge for larval culture to provide live food for at least two months after hatching. Post-metamorphosis juveniles, however, can be easily weaned to artificial diets.

Several more robust and fecund smelt species (compared to delta smelt) of the family Osmeridae have previously been cultured in North America, Japan, and Russia with limited success (Kendall 1926; Akielaszek et al., 1985; Moring, 1985; Kashiwagi et al., 1988; Ohama, 1990). In most cases, the culture strategy for smelts employed gamete stripping and artificial insemination followed by stocking embryos and newly hatched larvae into the natural watersheds and ponds. The rearing of larvae through metamorphosis and subsequent rearing of juveniles to sexual maturity have not been successful in the laboratory for any osmerid species; however, Mitsitano et al. (1977) have reported successful rearing of larvae of the Pacific surf smelt *Hypomesus pretiosus* to age 22 days using laboratory containers with cultured phytoplankton and rotifers.

Tentative and small scale delta smelt culture has been developed at UC–Davis using a temperature-controlled water recycling system with biological filtration (Mager et al., 1996). It includes the five basic steps illustrated in Figure 2. While these efforts resulted in the first documented success of breeding and rearing delta smelt through metamorphosis in the laboratory, the production characteristics of this culture system have not been adequate with regard to survival of juveniles and reproducibility of results. Major difficulties encountered included sensitivity of spawning adults and larvae to occasional elevation of ammonia and nitrites in the recirculated water, unstable spawning performance of brood fish in tanks with clean transparent water, and poor performance of the post-larvae during prolonged periods of feeding with brine shrimp nauplii.

Two research projects, lead by Dr. Serge Doroshov at UC-Davis and Dr. Joan Lindberg at SWP, were merged in 1997 under funding from the Interagency Ecological Program (Department of Water Resources with the Federal Bureau of Reclamation). We balanced the rearing program to take advantage of the delta canal-water source at the SWP for spawning most of the adults and rearing the post-larvae, and the clean well water at UC-Davis for incubating the eggs and rearing the early larvae. Additionally, the live food cultures required by the early larvae were established on clean well-water at Davis. Results of the 1997 culture efforts in the two water sources reveal: (1) delta smelt spawned better at the SWP site in natural canal water, (2) incubation of eggs proceeds better in clean well-water, (3) early-larval rearing results are not conclusive and tests of several water-rearing conditions are needed, and (4) post-larvae rear better in a flow-through water system using natural canal water compared to recirculation systems used at the UC–Davis laboratory.

A summary and interpretation of our 1997 results follows. Spawning success was markedly better at the SWP site than at UC-Davis. Of the 40,000 eggs produced from the two sites, the SWP site accounted for 81%. The delta-water source at the SWP site is more turbid and may reduce visual stressors, and it may also carry cues for spawning (e.g. odors, presence of phytoplankton or zooplankton, temperature cues). Incubation of the eggs proceeds better in well-water because the eggs are susceptible to fungal spores carried in delta water. Prophylactic treatment of the eggs with formaldehyde suppresses fungal infection in well-water, but fungal re-infection can occur from delta water laden with spores.

Over the last 2 years, the UC–Davis group has demonstrated success in rearing larval delta smelt, but losses were high and variable in 1997 (7-90%). Larvae were reared in static aquaria using clean well-water with and daily water replacement (75%). Rotifers were added each morning (5-12 per ml) followed by phytoplankton (*Nanochloropsis oculata*), which apparently stimulates the feeding response of larvae. Two inter-related factors appear to be responsible for the poor larval survival: the static-renewal method of water exchange, and the probable contamination of the larval tanks with a bacterial infection from the algal culture. If partially filtered canal water (containing some algae) promotes feeding in early larvae, this would eliminate the need to rear algae, and would decrease the disease risk to larvae as well. We plan to test this technique.

In 1997 we have reared 500 post-larval smelt to the 60 to 75 day-old-stage at the SWP site. Significant mortalities occurred this year after transfer of the post-larvae (30-day post-hatch larvae; 11 mm) from the UC-Davis site. The mortality rate was high for about 10 days following transfer of the fish (50% of the population), then mortalities diminished. The possible infection of larvae with bacteria from the algal culture may also have contributed to the high mortality rate. We plan a flow-through system for rearing the larvae, possibly eliminating the cultured algae, so that the larvae will be in better condition at the 30-day age. Since all rearing phases will be at one site, the larvae will not need to endure the extensive bagging and transfer procedures that were necessary in 1997 to move fish from UC–Davis to the SWP site. This should improve survival considerably. An increased zooplankton collection capacity in 1998 will also improve post-larval smelt growth and survival to metamorphosis.

Due to the importance of the flow-through system and delta-water source to successful smelt culture, we propose to continue using the existing small laboratory at the SWP site, and to use the new fish-holding facility at the nearby Federal Pumping site in place of the UC- Davis site. The Federal site has a temperature-controlled environment, and a clean well-water supply required for rearing the live-culture prey of the delta smelt larvae. Restricting the culture operation to one geographic area will minimize transport stress to the larvae, and provide economies in resources and labor.

In the proposed project, we will focus on improving fish culture techniques by testing factors influencing larval smelt growth and survival in the first two years, and establishing a final culture system in the third year. By the end of the third year we expect to produce cultured delta smelt at all life stages reliably. During the first two years we will be able to supply researchers with delta smelt embryos and yolk-sac larvae, and we may be able to supply small numbers of post-larvae and juveniles. In addition, we hope to obtain post-larvae from the pumping stations, either from the fish salvage tanks or from light traps.

During the last two years we have begun supplying smelt at various life stages to researchers. Healthy post-spawn individuals, have been supplied to Prof. Joe Cech, (UC–Davis), and Dr. Chuck Hanson (Hanson and Associates) for their research on swimming performance, fish screen design, and acoustical sensitivity of delta smelt. In 1997 we have preserved some embryo and larval fish samples for three projects: the embryo-biomarker work of Susan Anderson (UC-Berkeley); the comparative morphological study of delta smelt and wagasaki smelt at the early larval stage by Dr. Johnson Wang (consultant); and the larval otolith-aging work of Lenny Grimaldo (DWR) and Dale Sweetnam (Fish and Game). Requests for smelt at several life stages have been submitted to us for 1998. In future years, the demand for smelt may escalate significantly. The Federal Bureau of Reclamation is planning to build a new water diversion channel and fish screen for its Tracy site. They anticipate using delta smelt as a sensitive fish species for testing screen designs, requiring a large supply of delta smelt.

Culturing delta smelt is difficult due to the high sensitivity of adult smelt to stress, and the prolonged larval phase requiring live food. In spite of the difficulties, we have made significant progress. With a stable laboratory and well designed system for rearing and testing fish-culture methodology, we are confident we can achieve our objectives. Our project represents a cooperative effort between State and Federal agencies to contribute to the restoration of a threatened species.

III. e. Proposed Scope of Work

Considering the technical nature of the culture system required to rear delta smelt and the necessity to move the operation to a more favorable location, we propose a three-year study. Progress in the past has been hindered by lack of continuous support to carry through culture work from one year to the next. As our program progresses it is necessary to have year round support to raise a continuous supply of smelt at all life stages. The scope of work is outlined as three tasks, corresponding to the three years of the proposal.

Task 1: 1998

In the first year of the project we will need to make improvements to the existing laboratory at the SWP site. We will purchase equipment and construct our pilot-scale rearing operation for delta smelt and live prey culture at the new fishholding facility at the Federal site. This new building is about 1000 sq ft, of which a third is available for rearing delta smelt in 1998. The rest of the building will be used as a holding facility for salvaged fish for the Bureau of Reclamation.

We will dedicate the first year to a series of small scale experiments with larval fish rather than the pursuit of larger scale production techniques. This strategy will allow us to optimize rearing methodologies. During this year, we will be able to supply both embryos and yolk-sac larvae for research from captive fish, and perhaps some juvenile smelt from the post-larvae collected from fish salvage or from light traps.

<u>Physical improvements at the two sites</u>

Several improvements will be made to the SWP site to assure performance and security of the culture facility there. These include insulating the laboratory; building and installing a small heat exchanger to chill water inflow using drain-water outflow; and installation of a larger capacity, natural zooplankton collection system.

The Federal site has completed construction of a new building; we will design and build our own smelt rearing system in this building. We will need to install two spawning tanks (5' circular), an egg-incubation system, and a larval and juvenile fish-rearing system. The egg and larval system will be plumbed separately from the adult spawning tanks to minimize disease transfer. We will need to purchase and install temperature control equipment for these systems.

<u>Improving spawning performance of captive brood stock</u>

In the first year we will spawn the majority of the fish (300) at the SWP site in three 5' circular tanks using canal water. The canal water turbidity is high during the spawning season reducing visual disturbances. We will spawn a smaller population of fish (100) at the Federal site using well water to test that system (Feb.-June). We will minimize disturbances for the broodstock, keeping them shaded and dimly lit.

All eggs will be incubated in clean well water at the Federal site. Two types of egg incubators used in the past will be installed. Prophylactic treatment of the eggs (250 ppm formaldehyde) twice during the 9-day incubation period will be performed to prevent fungal infestations. This procedure should improve embryo survival substantially over the previous year in which 30% of the spawns were lost to fungus.

<u>Optimization of the larval culture procedures</u>

We suspect that rearing the larvae on filtered canal water (allowing some algae to remain) will promote feeding on rotifers. Two tanks (125-liter circular tanks) of larvae will be reared in this manner, and two tanks reared on well water with algae added.

Growth and survival in the larval tanks will be compared by sampling the tank population at 20 and 30 days post-hatch and recording daily mortalities.

In addition, we will conduct experiments to test for factors that influence early larvae feeding response. These will be bench top experiments with about 20 larvae in each of several 2-liter beakers. The measured response will be percent of fish with food in the gut. Factors to be tested include canal water and well water with and without algae.

Optimization of post-larval rearing through metamorphosis to sub-adults
Post-larvae, 20-30 days post-hatch, will be reared at both the federal and the SWP sites. Three tanks (125 liter) are available at the SWP site and 4 tanks at the federal site. Each tank can hold about 1500 post-larvae. We will also purchase two 3-ft diameter tanks for rearing the juvenile fish to the sub-adult stage.

We will improve the zooplankton collection system at the SWP site by making larger nets to handle a greater flow rate (200 gpm). We will be able to use the egg- and larval-sampling system for collecting zooplankton at the Federal site (200-gpm flow rate). In 1997, we used artemia nauplii to supplement the zooplankton diet; the larger zooplankton collection capacity should eliminate the need for artemia.

We will rear the post-larvae at gradually increasing temperatures (16-19 °C) over the spring and summer. The increase of 2-3 °C over 1997's rearing temperature should improve growth substantially given an ample supply of natural zooplankton. Growth of the larvae and juveniles will be measured by changes in mean length and dry weight. Stomach content analysis will be conducted on a sub-sample of the larvae and juveniles. Weaning of the juveniles to a commercial diet will be initiated at 60 days post-hatch. Transition to dry feed is expected at about 90 days post-hatch, based on 1997 data.

Capture of post-larvae from the field
In addition to rearing the post-larvae from the larval stage, we will obtain post-larval smelt (20-30 mm) from the fish salvage by skimming them off the top of the fish collection tanks, and by setting light traps. Fish collected from the wild will be held in slightly saline water for at least 24 hrs, and treated prophylactically with antibiotic. They will be reared in 2 tanks receiving canal water and fed natural zooplankton. Should this effort be successful, we may be able to greatly increase our supply of captive smelt for research in the subsequent two years.

Summary of data and preparation of report
Data on adult survival and spawns from the two sites will be compared. Egg fertility and larval hatch data from the two incubator types will be reported. Results of the short term rearing tests with the early larvae will be summarized and conclusions drawn. Survival data and measurement data for the post larvae will be summarized and graphed. Results of the two methods for collecting post-larvae from the wild will be discussed, and growth and survival data will be presented. The report will present conclusions as well as projections for the next year's work. The report will be delivered at the start of the 12th month.

Task 2: 1999
In the second year of our program, we will modify culture procedures based on 1998 results. In addition, we will test both larvae and the post-larvae for differential growth at two prey densities. We will install a temperature-control system to enable larval-rearing trials at three temperatures simultaneously. We will install six more larval tanks and two more juvenile tanks to expand our production of delta smelt and increase our supply of

live and preserved material to research laboratories. Our major focus will be on the optimization of temperature and feeding regimes and the preliminary evaluation of delta smelt performance in the production system (survival, development, and growth).

Task 3: 2000
In the third year of our program, we will make adjustments to our culture system based on previous years' results. We will characterize the final performance of smelt in the production culture system and evaluate the capability of this system to supply live material for research. We will summarize and publish the final methodology for delta smelt culture with recommendations for its application. The final report will be delivered at the start of the 36^{th} month.

References

Akielaszek, J. J., J. R. Moring, S. R. Chapman, and J. H. Dearborn. 1985. Experimental culture of young rainbow smelt, *Osmerus mordax*. Trans. Amer. Fish. Soc. 114: 596-603.

Kashiwagi, M., T. Iwai, and A. N. G. Lopes. 1988. Effects of temperature and salinity on egg hatch of the pond smelt, *Hypomesus transpacificus*. Bull. Fac. Bioresources, Mie University 1: 7-13.

Kendall, W. C. 1926. The Smelts. Bull. U.S. Bureau of Fisheries 42: 217-375.

Lindberg, J. C. 1992. Development of delta smelt culture techniques. Progress report, California Department of Water Resources.

Lindberg, J. C. 1996. Delta smelt culture, State Water Site, 1995. Final report, California Department of Water Resources, Contract B-59776, 24 pp.

Mager, R. C. 1996. Gametogenesis, reproduction, and artificial propagation of delta smelt, Hypomesus transpacificus. Ph.D. Dissertation, University of California–Davis, 125 pp.

Mager, R. C., S. I. Doroshov, and J. P. Van Eenennaam. 1996. Development of laboratory culture of delta smelt. Final report, Department of Water Resources, Fund title DWR B-59306, 65 pp.

Misitano, D. A. 1977. Technique for incubation and hatching eggs of surf smelt for bioassay. Prog. Fish. Cult. 39 (4): 187.

Moring, J. C. 1985. Smelt culture. Maine Fish and Wildlife Summer, pp 13-15.

Moyle, P. B., D. E. Stevens, and L. W. Miller. 1992. Life history and status of delta smelt in the Sacramento–San Joaquin Estuary, California. Trans. Amer. Fish. Soc. 121 (1): 67-77.

Ohama, H. 1990. Age determination of Japanese smelt with daily otolith increments. Nipp. Suis. Gak. 56 (7): 1053-1057.

Swanson, C. and J. J. Cech. 1995. Environmental tolerances and requirements of the delta smelt, *Hypomesus transpacificus*. Final report, California Department of Water Resources, Contracts B-59449 and B-58959, 77 pp.

Swanson, C., P. Young, and J. J. Cech. 1994. Delta smelt swimming performance, endurance, and behavior in a laboratory flume: preliminary results. 1994. Progress report, California Department of Water Resources.

U.S. Fish and Wildlife Service. 1995. Sacramento–San Joaquin Delta Native Fishes Recovery Plan. U.S. Fish and Wildlife Service, Portland, Oregon.

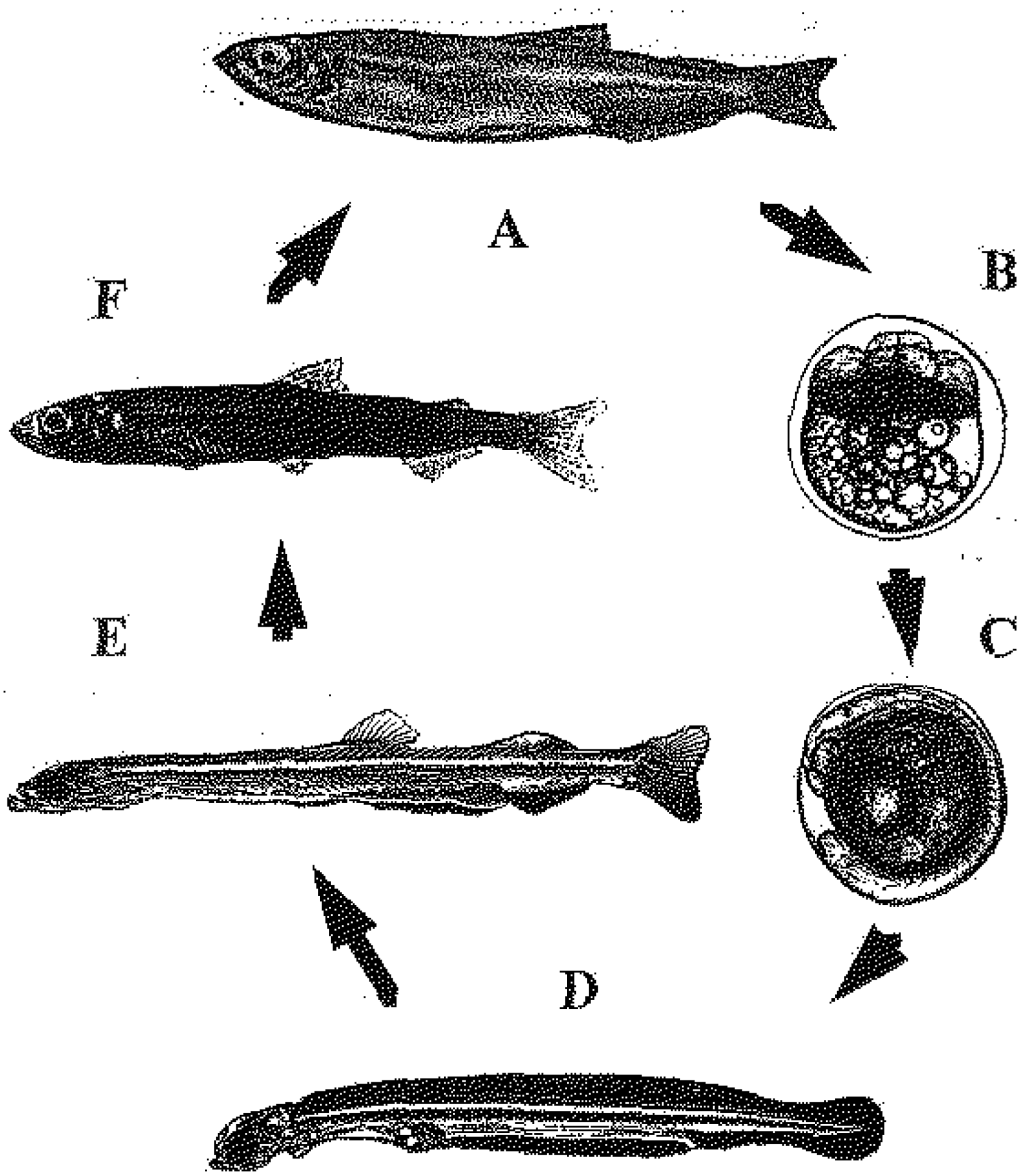

Figure 1

Life cycle of delta smelt (Mager, 1996). A - adult (length 70-80 mm). B - embryo (cleavage, egg diameter 1mm). C - embryo (organogenesis, 70 h post fertilization). D - prefeeding larva (5.5 mm, 3d post hatch). E - Feeding larva (differentiation of fins: 16mm, 40d post hatch). F - juvenile at metamorphosis (30mm, 100d post hatch).

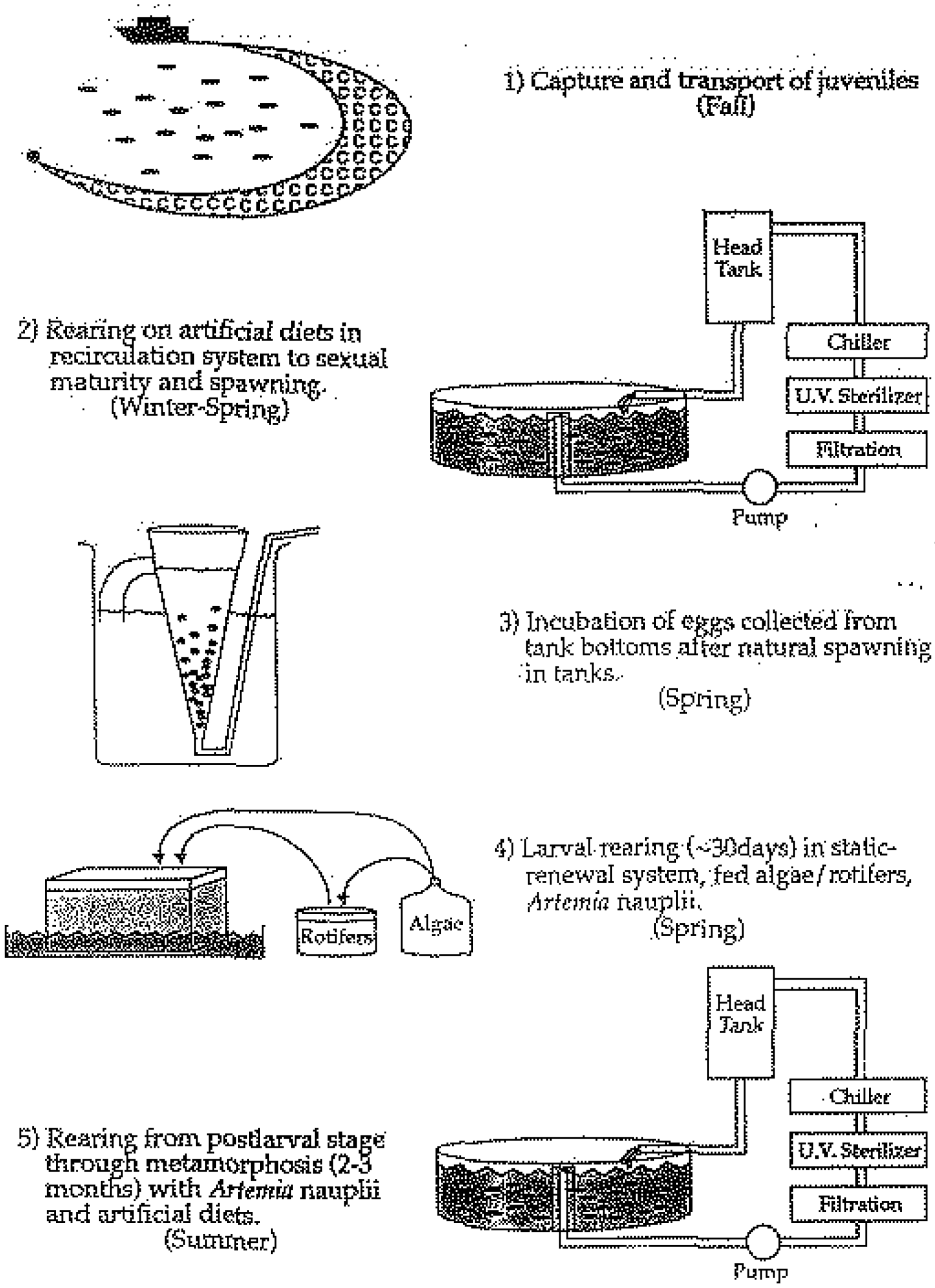

Figure 2

Laboratory culture system for delta smelt developed at UC Davis (Mager et al. 1996). Water recycling system for the steps 2 and 5 are being replaced with a flow-through system in our current project.

IV. a. Budget Costs

Personnel	Salary '98	Salary '99	Salary 2000	
Principal Investigator, Prof. Serge Doroshov	$0	$0	$0	
Project Administrator, Joel Van Eenennaam (Scientific Research Assoc., 2 mos.)	$8,620	$8,879	$9,234	
Project Manager, Dr. Joan Lindberg (Post Graduate Researcher, IX)	$41,500	$42,745	$44,455	
Brent Bridges (Post Graduate Researcher, III)	$32,500	$33,475	$34,814	
Jennie Kulczyk (Lab Assist, III)	$28,000	$28,840	$29,994	
Hourly Assist., TBN 1000 hrs. @ $10/hr	$10,000	$10,300	$10,712	
Subtotal Salary	**$120,620**	**$124,239**	**$129,209**	
Subtotal Benefits	**$21,398**	**$22,040**	**$22,922**	
Subtotal Salary and Benefits	**$142,018**	**$146,279**	**$152,131**	
Equipment	$9,500	$9,880	$10,275	
General Supplies	$18,000	$14,000	$14,000	
Travel	$8,500	$8,500	$8,500	
Total Direct Costs	**$178,018**	**$178,659**	**$184,906**	
Indirect Costs 10%, except equipment	$16,852	$16,878	$17,463	
Total Costs per Year	**$194,870**	**$195,537**	**$202,369**	
Grand, 3-year Total				**$592,776**

Budget Justification

Personnel: two full-time post-graduate researchers will manage the project, one will focus on broodstock, eggs and larvae, and one will focus on algae and rotifer production. One full time lab assistant will assist with all aspects of the project and the hourly employee will assist on weekends, evenings and daily routines during the spawning and larval rearing period. The part time research associate will be responsible for administration of the project at UC Davis.

Equipment: during the first year a water filtration unit ($1600), heat pump ($6400), salinity meter ($800) and heat exchanger ($700) are requested to upgrade culture capabilities at the State Water Projects fish facility. During the second year a spectrophotometer for water analysis ($2000), chiller/pump/filtration system ($7880) for a culture system at the Federal site. The third year equipment costs are estimated to be $10275 for continued expansion of the Federal site (pumps/chiller/filter/heat exchanger).

Supplies and Expenses: larval tanks and pvc ($1500), refractometer ($400), insulation for lab and tanks to maintain temperature ($2000), Delta water intake system ($2000), communication and alarm systems ($500), rotifer and algae supplies ($2500), culture system supplies ($3500), lab chemicals and glassware ($1000), water quality test kits and supplies ($1500), equipment parts and maintenance ($800), field sampling supplies ($1000), misc lab supplies ($800), xerox, computer and illustration services ($500). Similar costs are expected for years 2 and 3, primarily for the Federal site.

Travel: post graduate researcher travel between Davis and Byron ($6000), broodstock collection and trips to UC Davis ($1500), attend scientific meeting to present results ($1000).

IV. b. Schedule Milestones

The following milestones are based on a start date in the fall of 1997.

Task 1–1998

Build and install pilot culture system at Federal site. Completion: January, 1998.

Provide embryos and yolk sac larvae to research laboratories. Completion: April-July, 1998.

Test effect of algae on larval feeding response. Completion: August, 1998.

Year end report. Completion: December, 1998.

Task 2–1999

Enlarge smelt culture system at the Federal site. Completion: January, 1999.

Provide embryos and yolk sac larvae to research laboratories. Completion: April–July, 1999.

Test effect of temperature on survival and growth of larvae and juveniles. Completion: September, 1999.

Task 3–2000

Spawn captive smelt. Completion: June, 2000.

Provide post-larvae and juveniles to research laboratories. Completion: June–September, 2000.

Deliver 3-year final report. Completion: December, 2000.

V. Applicant Qualifications

Dr. Serge Doroshov, Principal Investigator

Education

Ph.D.: Biology/Oceanography, Academy of Science, Moscow, Russia, 1967.
M.S. and B.S.: Zoology/Icthyology, University of Moscow, Russia, 1959.

Employment History

1995-present: Director of the Aquaculture and Fisheries Program, University of California–Davis.
1978-present: Associate Professor and Professor of Animal Science, University of California–Davis.
1967-1975: Head of the Laboratory of Mariculture, VNIRO, Moscow, Russia.

Research Experience

Developmental biology and reproductive physiology of fish (striped bass, sturgeon, delta smelt, catfish, trout). Fish culture and hatchery technology.

Dr. Joan C. Lindberg, Project Manager

Education

Ph.D.: Ecology, University of California–Davis, 1988. Dissertation: Feeding and behavior studies in larval and juvenile white sturgeon, *Acipenser transmontanus*.
M.S.: Zoology, University of Wisconsin–Madison, 1983.
B.S.: Zoology, University of Wisconsin–Madison, 1979.

Employment History

1996-1997: Postgraduate researcher, University of California–Davis.
1994-1996: Research associate, San Francisco State University, San Francisco, CA.
1990-1994: Fish biologist, BioSystems Analysis, Tiburon, CA.
1990: Instructor of General Biology, Las Positas College, Livermore, CA.
1988-1990: Postdoctoral study, Lawrence Livermore National Lab, Livermore, CA.

Research Experience

Project development, management and construction of of delta smelt culture program. Design and implementation of pilot study to assess use of restored wetland habitat for spawning by delta smelt. Research on salmon imprinting physiology, research on juvenile sturgeon feeding behavior in culture system. Assessment of molecular toxicology technique to detect DNA damage in striped bass.

Joel Van Eenennaam, Project Administrator

Education

MS: International Agriculture Development (Aquaculture Specialization), University of California–Davis, 1985.
BS: Fisheries and Wildlife, Michigan State University, 1977.

Employment History
1985-present: Research Associate, UC Davis.
1983-1985: Research Assistant, UC–Davis
1982: AquacultureTechnician, Fish Breeders of California.
1977-1981: Fisheries Extension Agent, Khon Kaen, Thailand.

Research Experience
Reproductive and developmental biology of cultured fish (sturgeon, paddlefish, striped bass, catfish, trout, common, chinese and indian carps, tilapia, bluegill). Development of hatchery technolgy in aquaculture. Organization of workshops in broodstock development, spawning induction, egg and larval rearing. Supervision of the sturgeon broodstock development program in California and the western region. Research on the reproductive conditions of Atlantic sturgeon on the Hudson River, NY. Supervision of several wet and dry laboratories at UC–Davis for research on reproductive biology of fish.

Brent Baskerville-Bridges, Post-Graduate Researcher
Education
M.S.: Ecology, University of California–Davis, 1996.
B.S.: Aquaculture and Fisheries, University of California–Davis, 1990.

Employment History
1996-Present: Post-Graduate Researcher, University of California–Davis.
1995: Post-Graduate Researcher, University of California–Davis.
1991-1993: Aquaculture and Fisheries Biologist, Tenera, Berkeley, CA.
1991: Hanson Environmental, Walnut Creek, CA.
1987-1990: Undergraduate Researcher, University of California–Davis.

Research Experience
Researched the potential role of bacterial particles as nutrition for larval fish. Designed, built, and managed a small-scale aquaculture holding facility for striped bass. Installed and maintained remote wilderness temperature recording stations. Designed and installed a raw water intake system in the Sacramento/San Joaquin Delta. Researched, designed, and built facility to culture microalgae and rotifers for delta smelt culture.

Jennie Kulczyk, Laboratory Assistant
Education
B.S.: Biology, Earlham College, Richmond, Indiana, 1995

Employment History
1997-present: Laboratory assistant, University of California–Davis.
1996-1997: Seasonal aid, California Department of Fish and Game, Stockton, CA.

Research Experience
Spawning and rearing of embryos, larvae, and post-larval smelt.

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DEPARTMENT OF WILDLIFE, FISH, AND CONSERVATION BIOLOGY
DAVIS, CALIFORNIA 95616-8751
FAX: (916) 752-4154

7-26-97

CALFED Proposal Reviewers
Sacramento, CA

Dear Reviewers:

I enthusiastically support the delta smelt rearing proposal of Dr. Doroshov and Dr. Lindberg. They plan to test a variety of factors considered important in the rearing success of this fish. In small scale experiments, they will examine effects of water chemistry and the presence of algae on the rotifer feeding response in the youngest (feeding) life history stages of delta smelt. They also plan to test temperature effects on larval and juvenile growth. These experiments should lead to improved delta smelt production capabilities by the third year of the study.

Many of us recognize the need for delta smelt. The reared fish would be very useful for toxicity testing, for experiments with fish screening devices (such as those that my colleagues and I have proposed to CALFED), and potentially for testing habitat suitability or for restocking parts of the Sacramento - San Joaquin Delta estuary.

The proposed project would be a cooperative effort between California and federal agencies to surmount the final hurdles regarding the mass production of delta smelt. I fully support this effort.

Sincerely,

Joseph J. Cech, Jr.
Professor of Fisheries Biology

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Dr. Serge I. Doroshov
Director of Aquaculture and Fisheries
Department of Animal Sciences
3202 Meyer Hall, University of California at Davis
Davis, California 95616
Tel: (916) 752-7603; Fax: 752-4508
e-mail: sidoroshov@ucdavis.edu

RE: Calfed grant proposal

Dear Dr. Doroshov,

We were delighted to learn that you are submitting a proposal to Calfed on laboratory breeding and culture of delta smelt for developmental and environmental studies. As stated in our proposals to Calfed (Bennett, Teh , and Anderson) and IEP (Teh and Hinton), we are requiring laboratory-raised delta smelt for ambient water toxicity testing and for determination of responses to exposure. We will compare the laboratory-exposed fish to field-collected fish and correlate cause and effect of contaminants in the Delta ecosystem. Furthermore, due to the fact that there is no delta smelt control site in California, your proposal is very critical to all investigators working on contaminant effects in delta smelt. The funding of your study will provide a unique interdisciplinary, lab-field approach not otherwise available.
We strongly support your proposal and will be glad to collaborate with you on your developmental studies.

Sincerely,
David E. Hinton Swee Teh

Swee J. Teh and David E. Hinton (Director)
Aquatic Toxicology Program
University of California-Davis